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MRID No. ~~427741-05~~

DATA EVALUATION RECORD

- 1. **CHEMICAL:** Dicamba.  
Shaughnessey No. 029801.
- 2. **TEST MATERIAL:** Dicamba technical; Lot No. 52103810; 86.9% purity; an off-white chunky solid.
- 3. **STUDY TYPE:** 71-1A. Avian Single Dose Oral LD<sub>50</sub> Test.  
Species Tested: Bobwhite quail (*Colinus virginianus*).
- 4. **CITATION:** Campbell, S.M., J. Grimes, M. Jaber, and J.B. Beavers. 1993. Technical Dicamba: An Acute Oral Toxicity Study with the Northern Bobwhite. Project No. 131-179A. Performed by Wildlife International Ltd., Easton, MD. Submitted by Sandoz Agro, Des Plaines, IL. EPA MRID No. ~~427741-05~~. 42918001. m)

5. **REVIEWED BY:**

Mark A. Mossler, M.S.  
Associate Scientist  
KBN Engineering and  
Applied Sciences, Inc.

Signature: *Mark A. Mossler*  
Date: 7/26/93

6. **APPROVED BY:**

Michael L. Whitten, M.S.  
Wildlife Toxicologist  
KBN Engineering and  
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Signature: *Michael L. Whitten*  
Date: 7/26/93  
*Abbas A. Faruqi* 10/28/93

Henry T. Craven, M.S.  
Supervisor, EEB/EFED  
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Signature: *Henry T. Craven*  
Date: 11/2/93

- 7. **CONCLUSIONS:** This study is scientifically sound and fulfills the requirements for an acute oral toxicity test using the bobwhite quail. The LD<sub>50</sub> value of 216 mg/kg (188 mg ai/kg) classifies technical dicamba as moderately toxic to bobwhite quail. The NOEL was 15.6 mg/kg (13.6 mg ai/kg).
- 8. **RECOMMENDATIONS:** N/A.
- 9. **BACKGROUND:**
- 10. **DISCUSSION OF INDIVIDUAL TESTS:** N/A.



**11. MATERIALS AND METHODS:**

- A. **Test Animals:** The birds used in the study were 26-week-old bobwhite quail (*Colinus virginianus*) obtained from a supplier in Belvidere, NJ. The birds were from the same hatch, pen-reared, and phenotypically indistinguishable from wild birds. They were acclimated to the laboratory for 12 weeks prior to testing and ranged in weight from 178 to 228 g at test initiation. Except for a 15-hour fasting period immediately prior to dosing, water and a game bird ration were offered *ad libitum* during acclimation and testing. No antibiotics were administered during the test.
- B. **Test System:** All birds were housed indoors in pens constructed of wire grid and steel sheeting. The pen floors measured 78 x 51 cm and the ceiling height ranged from 20-25 cm. Fluorescent lights provided 8 hours of 285 lux illumination per day. The average temperature was  $19.2 \pm 1.8^{\circ}\text{C}$  and the average relative humidity was  $50 \pm 9\%$ .
- C. **Dosage:** Fourteen-day single dose oral LD<sub>50</sub> test. Based on known toxicity data, seven nominal dosages (15.6, 31.2, 62.5, 125, 250, 500, and 1000 mg/kg of body weight) and a vehicle (corn oil) control were selected for the test. The dosages were not corrected for the percent active ingredient (ai) of the test substance.
- D. **Design:** Groups of ten birds (five males and five females) were assigned to each treatment and control group by indiscriminate draw. Each dosage group was assigned two pens in which the birds were segregated by sex.

The test substance was dispersed in corn oil. Each bird was individually weighed and dosed on the basis of milligrams of test substance per kilogram of body weight. The dose was administered with a stainless steel cannula in the crop or proventriculus of the birds. The control birds received corn oil only. All birds were dosed at a constant volume of 6 ml per kilogram of body weight.

All birds were observed once a day during acclimation and at least twice daily during testing for mortality, signs of toxicity, and abnormal behavior. The birds were individually weighed at test initiation and by

group on days 3, 7, and 14. Group food consumption was determined for days 0-3, 4-7, and 8-14 by measuring the change in feed presented to the birds over a period of time. However, this is an estimate due to wastage by the birds.

E. **Statistics:** Mortality data were analyzed with the use of a computer program that employed three methods of analysis (probit analysis, moving average angle method, and binomial probability method). The probit analysis method was used in this study to determine the LD<sub>50</sub>.

12. **REPORTED RESULTS:** There were no mortalities in the control, 15.6, 31.2, or 62.5 mg/kg treatment groups. Ten percent mortality was noted in the 125 mg/kg group, 60% occurred at the 250 mg/kg level, and 100% mortality was observed for the 500 and 1000 mg/kg groups. All birds in the control and 15.6 mg/kg groups were normal in appearance and behavior.

At the 31.2 mg/kg level, signs of toxicity were first exhibited in two females approximately 30 minutes after dosing and continued through the morning of day 4. All other birds were normal in appearance and behavior. Signs of toxicity were lower limb weakness, lower limb rigidity, wing droop, loss of coordination, reduced reaction to external stimuli (sound and motion), a ruffled appearance, lethargy, and stiff walking.

At the 62.5 mg/kg level, signs of toxicity were first exhibited 30 minutes after dosing and continued through the afternoon of day 5. From the morning of day 6 until study termination, all birds were normal in appearance and behavior. Signs of toxicity were lower limb weakness, wing droop, loss of coordination, loss of righting reflex, reduced reaction to external stimuli, lethargy, a prostrate posture, crouching, and a ruffled appearance.

At the 125 mg/kg level, signs of toxicity were first exhibited 20 minutes after dosing and continued through the afternoon of day 5. The single mortality was noted on day 1. From the morning of day 6 until study termination, all surviving birds were normal in appearance and behavior. Signs of toxicity were shallow and rapid respiration, lower limb weakness, lower limb rigidity, wing droop, reduced reaction to external stimuli, wing flapping, lethargy, a ruffled appearance, loss of righting reflex and coordination, walking stiffly, a prostrate posture, depression, and coma.

At the 250 mg/kg level, signs of toxicity were first exhibited 15 minutes after dosing and two birds were found dead within one hour. Additional mortalities were noted on the mornings of days 1, 2, and 3. From the morning of day 5 until study termination, all surviving birds were normal in appearance and behavior. Signs of toxicity were shallow and rapid respiration, lower limb weakness, lower limb rigidity, wing droop, reduced reaction to external stimuli, loss of coordination and righting reflex, wing flapping, a ruffled appearance, depression, and a prostrate posture.

At the 500 mg/kg level, signs of toxicity were first exhibited 10 minutes after dosing. By the end of day 0, six birds had died, with the first mortality occurring within one hour after dosing. Two birds were found dead on day 1, and the two remaining birds were found dead on day 3. Signs of toxicity were shallow and rapid respiration, gasping, lower limb rigidity, wing droop, reduced reaction to external stimuli, loss of righting reflex, prostrate posture, ruffled appearance, wing flapping, and depression.

At the 1000 mg/kg level, signs of toxicity were first exhibited immediately after dosing. By the end of day 0, eight birds had died, with the first mortality occurring within 45 minutes after dosing. By the morning of day 1, all birds had died. Signs of toxicity were lower limb rigidity, wing droop, reduced reaction to external stimuli, loss of coordination, prostrate posture, loss of righting reflex, wing flapping, and gasping.

There was a reduction in body weight gain among males and females at the 31.2 and 62.5 mg/kg levels during days 0-3. A loss of body weight was noted for all birds in the 125 and 250 mg/kg groups. There was a reduction in feed consumption at the 125 and 250 mg/kg levels during days 0-3 (Table 2, attached). Feed consumption and body weight trends could not be determined for the two highest levels, due to total mortality.

13. **STUDY AUTHOR'S CONCLUSIONS/QUALITY ASSURANCE MEASURES:**

The acute oral LD<sub>50</sub> value for bobwhite quail exposed to technical dicamba was determined to be 216 mg/kg, with a 95% confidence interval of 162-288 mg/kg. The slope of the probit curve was 6. The no mortality dosage was 62.5 mg/kg. The no-observed-effect level (NOEL) was 15.6 mg/kg, based on signs of toxicity and reductions in body weight gain at the 31.2 mg/kg level.

Quality Assurance and Good Laboratory Practice (GLP) statements were included in the report indicating compliance

with the regulations set forth in 40 CFR Part 160 with the following exception: samples of the dosing solutions were not collected for dose confirmation, stability, or homogeneity analyses. Additionally, it was not known if the test substance characterization was performed under GLP standards.

14. REVIEWER'S DISCUSSION AND INTERPRETATION OF STUDY RESULTS:

A. Test Procedure: The test procedures were in accordance with Subdivision E and SEP guidelines with the following exception:

Group body weights, rather than individual body weights, were taken on days 3, 7, and 14.

Necropsies were not conducted on the test birds. These are recommended, but not required.

B. Statistical Analysis: The reviewer used EPA's Toxanal program to verify the LD<sub>50</sub>. The results are the same as those of the authors (see attached printout).

C. Discussion/Results: This study is scientifically sound and fulfills the requirements for an acute oral toxicity test using the bobwhite quail. The LD<sub>50</sub> value of 216 mg/kg (188 mg ai/kg) classifies technical dicamba as moderately toxic to bobwhite quail. The NOEL was 15.6 mg/kg (13.6 mg ai/kg).

D. Adequacy of the Study:

(1) Classification: Core.

(2) Rationale: N/A.

(3) Repairability: N/A.

15. COMPLETION OF ONE-LINER: Yes, 7-24-93.

MOSSLER DICAMBA COLINUS VIRGINIANUS 7-24-93

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CONC.	NUMBER EXPOSED	NUMBER DEAD	PERCENT DEAD	BINOMIAL PROB. (PERCENT)
1000	10	10	100	9.765625E-02
500	10	10	100	9.765625E-02
250	10	6	60.00001	37.69531
125	10	1	10	1.074219
62.5	10	0	0	9.765625E-02
31.2	10	0	0	9.765625E-02
15.6	10	0	0	9.765625E-02

THE BINOMIAL TEST SHOWS THAT 125 AND 500 CAN BE USED AS STATISTICALLY SOUND CONSERVATIVE 95 PERCENT CONFIDENCE LIMITS, BECAUSE THE ACTUAL CONFIDENCE LEVEL ASSOCIATED WITH THESE LIMITS IS GREATER THAN 95 PERCENT.

AN APPROXIMATE LC50 FOR THIS SET OF DATA IS 220.3337

RESULTS CALCULATED USING THE MOVING AVERAGE METHOD

SPAN	G	LC50	95 PERCENT CONFIDENCE LIMITS	
5	.1144044	210.6757	147.4024	315.0845

RESULTS CALCULATED USING THE PROBIT METHOD

ITERATIONS	G	H	GOODNESS OF FIT PROBABILITY
7	.3122431	1	.9972041

SLOPE = 5.960624  
95 PERCENT CONFIDENCE LIMITS = 2.629904 AND 9.291344

LC50 = 215.7824  
95 PERCENT CONFIDENCE LIMITS = 161.9506 AND 287.9937

LC10 = 132.1146  
95 PERCENT CONFIDENCE LIMITS = 64.8029 AND 172.8131

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TABLE 2  
AVERAGE BODY WEIGHT AND ESTIMATED FEED CONSUMPTION OF NORTHERN BOBWHITE  
GAVAGED WITH TECHNICAL DICAMBA

Dosage mg/kg	Sex	Average Body Weight in Grams								Estimated Feed Consumption Grams/Bird/Day		
		Day 0	Change	Day 3	Change	Day 7	Change	Day 14	Total Change	Days 0-3	Days 4-7	Days 8-14
Control	M	201	7	208	-1	207	2	209	8	17	21	19
	F	197	3	200	1	201	-3	198	1	17	18	19
15.6	M	190	7	197	1	198	0	198	8	15	19	16
	F	210	10	220	-1	219	-3	216	6	23	25	22
31.2	M	215	4	219	0	219	1	220	5	15	19	20
	F	207	1	208	1	209	0	209	2	20	23	23
62.5	M	205	3	208	-4	204	-2	202	-3	26	25	23
	F	208	0	208	-1	207	2	209	1	24	24	22
125	M	201	-11	190	5	195	6	201	0	12	38	33
	F	199	-1	198	1	199	4	203	4	11	22	23
250	M	199	-9	190	7	197	4	201	2	10	26	25
	F	201	-7	194	-1	193	4	197	-4	4	45	32
500	M	201	-	-	-	-	-	-	-	-	-	-
	F	204	-	-	-	-	-	-	-	-	-	-
1000	M	212	-	-	-	-	-	-	-	-	-	-
	F	211	-	-	-	-	-	-	-	-	-	-

(-) Data not available due to total mortality.